Topic 12 – Myocardial hypoxia, reperfusion, stroke – A

April 02nd, Thursday 2015

0192

Pharmacological modulation of TRPV1, a cation leak channel in mouse cardiomyocytes

Ribal Al-Mawla, Lucille Païta, Sylvie Ducreux, Christophe Chouabe, Michel Ovize, Fabien Van Coppenolle
Faculté de Médecine, INSERM1060, Lyon, France

The sarcoplasmic reticulum (SR) calcium homeostasis is due to a dynamic balance between the capture of the cytosolic calcium by calcium pumps such as SERCA (Sarco Endoplasmic Reticulum Calcium ATPase) and calcium release, actively across calcium channels such as IP3 (Inositol Tri-phosphate) and ryanodine receptors, or passively via calcium leak channels.

Few data are available concerning the functional characterization of leak channels in the SR. These channels are involved in the regulation of the reticular calcium concentration as well as in the exchange of calcium between SR and other intracellular organelles. Therefore, their characterization is important for a better understanding of the physiology of the cells.

Recently, we have demonstrated that TRPV1 (Transient Receptor Potential Vanilloid 1), a cationic channel, is a functional calcium leak channel on the SR of mouse skeletal muscle cells. TRPV1 is activated by acidosis, high temperature (>42°C), and by pharmacological molecules such as capsaicin, resiniferatoxin and capsazepin.

We are pursuing our investigation of these channels in C57Bl6 mouse cardiomyocytes.

Our preliminary results show that TRPV1 is active in mice cardiomyocytes as a calcium leak channel after stimulation or inhibition using pharmacological molecules.

These data were confirmed by using a genetic approach: the C57Bl6 KO mice for TRPV1 channel.

Lately, reducing cardiac injuries after ischemia reperfusion where calcium dynamics play a crucial role became a major interest. Therefore, the modulation of the TRPV1 calcium leak channel might be a new approach in cardioprotection.

0227

Mineralocorticoid receptor deficiency in smooth muscle cells protects against renal injury induced by ischemia/reperfusion

Jonatan Barrera-Chimal (1), Sonia Prince (1), Fouad Fadel (1), Alan Le Mercier (1), Soumaya El Moghrabi (1), Norma Bobadilla (2), Frederic Jaisser (1)
(1) Centre de Recherche des Cordeliers, INSERM U1138 Team 1, Paris, France – (2) Instituto de Investigaciones Biomedicas, Molecular Physiology, UNAM and INNSZ, Mexico City, Mexique

Introduction: Renal ischemia/reperfusion (IR) is an important cause of acute kidney injury (AKI). AKI has been linked with progression to chronic kidney disease and development of cardiac alterations. Previous studies from our laboratory have shown that mineralocorticoid receptor (MR) antagonism with spironolactone prevents tubular injury and renal dysfunction induced by IR. The protection mechanisms remain unclear and whether the blockade of vascular MR is responsible for the protection conferred by MR antagonists in IR remains unexplored.

Objective: To evaluate the specific contribution of vascular MR in the development of the kidney injury induced by IR.

Methods: To investigate the contribution of vascular MR we generated two knockout (KO) mouse models. To allow MR inactivation in endothelial cells (MR^{endoKO} mice), floxed MR mice ($MR^{fl/fl}$) were crossed with mice expressing the Cre recombinase under the Tie2cre promoter. To allow MR inactivation in smooth muscle cells (MR^{SMCKO} mice), $MR^{fl/fl}$ mice were crossed with mice expressing the inducible Cre recombinase under the SMA promoter (MR^{SMCKO}). In these mice, sham surgery or bilateral renal IR for 20min was performed. The animals were studied 24h after reperfusion. As markers of tubular injury, the mRNA levels of Kim-1 and NGAL were quantified.

Results: In $MR^{fl/fl}$ mice, IR induced renal dysfunction (plasma creatinine raised from 8.9±0.3 in sham to 33.8±4.8 µmol/L in IR), tubular injury and an increase in mRNA levels of Kim-1 (400-fold) and NGAL (200-fold). The MR^{endoKO} mice displayed similar alterations induced by IR as $MR^{fl/fl}$ mice. In contrast, after renal IR, the MR^{SMCKO} mice presented normal renal function (plasma creatinine was 9.6±0.7 and 14.0±1.9 µmol/L in sham and IR, respectively), absence of histological alterations and a significant reduction in the mRNA levels of Kim-1 and NGAL. We are now testing the effect of MR deficiency in smooth muscle cells in preventing the development of chronic alterations induced by IR.

Conclusion: We show that the deficiency of the MR expressed in smooth muscle cells protects against the renal injury induced by IR. Our data provides further evidence to support the use MR antagonists as a novel therapeutic approach to prevent acute and chronic consequences of renal IR.

0374

Cardioprotection against ischemia-reperfusion injury by heart rate control

Delgado Viviana (1), Aurelie Covinhes (1), Pietro Mesirca (1), Isabelle Bidaud (1), Joel Nargeot (2), Christophe Piot (2), Jorg Striessnig (3), Matteo Mangoni (1), Stéphanie Barrère-Lemaire (4)
(1) Institut de Génomique Fonctionnelle, INSERM U661 – CNRS UMR5203 – UM1, UM2, Montpellier, France – (2) Institut de Génomique Fonctionnelle, INSERM U661 – CNRS UMR 5203 – UM1, UM2 – Physiologie, Montpellier, France – (3) University of Innsbruck, Center for Molecular Biosciences, Innsbruck, Autriche – (4) Institut de Génomique Fonctionnelle, Cardioprotection, Montpellier, France

Background: Acute myocardial infarction (AMI) is a major cause of mortality worldwide. Early reperfusion is the only treatment recommended to reduce infarct size. However, reperfusion induces also deleterious secondary effects called ischemia-reperfusion (IR) injury due to irreversible apoptotic death of cardiomyocytes. Most ischemic episodes are triggered by an increase in heart rate that induces an imbalance between myocardial oxygen delivery and consumption. The BEAUTIFUL clinical trial has demonstrated that moderate heart rate reduction diminishes the frequency of AMI episodes in patients with stable coronary artery disease having increased heart rate at rest. The HCN-mediated I_f current and the $Ca_v1.3$-mediated L-type Ca^{2+} currents play important roles in the generation of automaticity and heart rate, therefore they are interesting targets for selective control of heart rate and cardioprotection during AMI. The aim of this study was to investigate if $Ca_v1.3$ channels could be a putative target to reduce infarct size.

Methods: Anesthetized C57BL/6J, $Ca_v1.3^{-/-}$ and $Girk4^{-/-}$ mice were subjected to a surgical protocol of myocardial IR (40min ischemia-60 min reperfusion). Heart rate was measured with a one-lead surface ECG recording, and infarct size with triphenyl tetrazolium chloride staining.

Results: Selective heart rate decrease (26%) in an *in vivo* mouse model of AMI is associated with reduced IR injury. Ivabradine administration before ischemia significantly reduced infarct size (–33%). $Ca_v1.3^{-/-}$ mice presented reduced infarct size (–30%) compared to WT mice. In addition, $Girk4^{-/-}$ mice, a genetic model of moderate tachycardia (10%) displayed increased infarct size (+30%) compared to control mice.

Conclusions: These results show a direct relationship between heart rate and IR injury. Heart rate reduction by inhibition of $Ca_v1.3$ channels constitutes a promising strategy to reduce infarct size.